73. (New) A method according to claim 72, wherein the first molecule or first molecular complex is positioned by chemical binding, adhesion, or condensation reactions.--

Remarks

Applicants thank Examiner Sisson for the courtesy of an interview on July 11, 2002.

Claims 21-25, 27-34, 37-40, 42-55 and 59-62 are currently pending. Herewith Applicants cancel claims 21, 22, 47, 48, 49, 51, 52, 55, 60 and 61, without prejudice or disclaimer. Applicants further add new claims 63-73. Thus, with the entry of this amendment, claims 23-25, 27-34, 37-40, 42-46, 50, 53-54, 59, and 62-73 will be active in this case. No new matter is added with this amendment or new claims.

Support for new claim 63 can be found at page 3, line 26 and page 5, line 2 ("equal or less than 1 μm"); at page 5, line 25 to page 6, line 4 ("sample"); page 3, lines 16-21 ("alternating current"); page 6, line 26 to page 7, line 3 ("peptides and proteins"); page 3 ("wherein the electrode structures are arranged so closely...").

I. Claim Objections

In paragraph 3, the Examiner objects to claims 52 and 55. Specifically, the Examiner states that both claims 52 and 21 stipulate that the electrode structures are insulated from one another. He also states that claim 55 indicates that direct or alternating current is to be applied yet claim 21 stipulates that an alternating electric field is generated. Thus, according to the Examiner, claim 55 broadens the scope of the invention. In response, Applicants point out that all of the objected to claims have been canceled. The new claims do not invoke these alleged inconsistencies.

II. Rejections under 35 USC § 112

The Examiner rejects claims 21-25, 27-34, 37-40, 42-55 and 59-62 for lack of enablement. Applicants traverse this rejection as it may be applicable to the remaining and new claims.

The Examiner argues that Applicants only exemplify the detection of one molecule and concludes that the specification does not provide enablement for the full scope of the claimed invention. Applicants respectfully traverse this rejection and direct the Examiner's attention to the Rule 132 declarations already of record in this case. (See Applicants response of August 27, 2001 and attached Exhibits.)

The Examiner also objects to claim 23 for reciting "impedance spectroscopy". In response, Applicants urge that impedance spectroscopy is an old and well know measurement technique.

For example, http://www.corrosion-doctors.org/Electrochem/EIS.htm states that "EIS has been successfully applied to the study of corrosion systems for thirty years." More than 250 issued U.S. Patents mention impedance spectroscopy, many in the molecular biology area. For example, U.S. Pat. No. 6,455,243 (priority date 6/12/98), titled "Nutritional assessment by measuring mitochondrial complex activity" cites a 1994 article by Schmuckler, "Impedance Spectroscopy of biological cells," Engineering in Medicine and Biology Society, 1994. Engineering Advances: New Opportunities for Biomedical Engineers, Proceedings of the 16th Annual Internal Conference of the IEEE. U.S. Pat. No. 6,297,059, "Triggered optical biosensor" Priority, 6/98, cites a 1993 article by Terrettaz et al. (1993). Protein binding to supported lipid membranes: investigation of the cholera toxin-ganglioside interaction by simultaneous impedance spectroscopy and surface plasmon resonance. Langmuir. 9:1361-1369 (also cited in U.S. Pat. No. 6,087,182, "Reagentless analysis of biological samples" Filed Aug. 27, 1998). U.S. Pat. No. 6,263,294, "Impedance spectroscopy measurement system," Filed March, 1998, cites to Kendig, Martin W., "Overview of the Science and Capability of Impedance

Spectroscopy" (Date Unknown). The specification itself refers to the work of Knichel et al. at page 2, lines 1 and 2. (These patents and articles are not being cited as relevant prior art, only to show that one of skill in the art would know how to perform impedance spectroscopy.)

The Examiner further asserts that the specification does not enable the application of "a direct-current component," (present claim 27 and 28) nor the oxidation or reduction of an electrically active molecule (claim 29). With regard to the "direct-current component" issue, Applicants point out that the specification at page 3, lines 16-21, discloses that the basic current must be an alternating current, *i.e.*, a current resulting from an alternating field. However, Applicants understood and considered as part of their invention, the fact that direct-current components can be superimposed over an alternating current. See the specification at page 7, lines 5 to 9 and the attached article "About Photovoltaics" which discusses these basic principles and adding ac and dc components in the context of photovoltaics. Because these principles are, in fact, known to the skilled artisan, Applicants respectfully request the Examiner to withdraw this rejection.

With regard to the "oxidation-reduction" issue, Applicants assert that the Examiner has not provided any reason to doubt Applicants' recited method. It is the Examiner's burden to set forth evidence as to why one of skill in the art would not believe the Applicants' assertions.

At page 6, the Examiner states that the specification has not set forth a sufficient number of members of the genus of electrochemical reaction to enable the genus, to enable the full scope of the invention, to detect an antibody (claim 40), to detect polynucleotides (claims 42-44) or to use a material other than gold.

In response to these comments, Applicants again direct the Examiner's attention to the Rule 132 Declaration and related data submitted to the Examiner on December 29, 2000 and the Declaration of Dr. Hintsche dated August 9, 2001. These declarations

presented post-filing evidence that by using the methods taught in the specification, Applicants were able to successfully apply their invention to the detection of a variety of molecules. In these declarations, the inventors also pointed out that all of the tools employed in their invention and disclosed in their specification were known and well accepted to the skilled artisan at the time of the invention. In view of these declarations and supporting documentation, Applicants respectfully request Examiner Sisson to reconsider his position and withdraw this rejection.

The Examiner further alleges at page 6 that claim 21 recites the limitation that the molecule or molecular complex comprises a nucleic acid or antibody but the claim does not necessarily require that the detection be that of the nucleic acid or antibody but rather, the detection could be directed to some other molecule that is present. Applicants respond by noting that new claim 63, part (d), addresses the Examiner's concern. Withdrawal of this rejection is therefore requested.

The Examiner also asserts that the claims have sufficient breadth of scope so as to encompass the use of electrodes that are in contact with one another. Applicants traverse this rejection and assert that new claim 63 clarifies this issue. Withdrawal of this rejection is therefore requested.

At page 7, the Examiner questions the meaning of "self-assembly" in claim 33. Applicants respond by pointing to page 7, line 19 to page 8, line 2 wherein this process is discussed in context with binding mechanisms. In any event, the amendment to claim 31 should resolve any confusion about dependent claim 33.

In paragraph 6 *et seq.*, the Examiner raises numerous rejections for indefiniteness. One issue relates to the heterogeneity of the sample. Applicants respectfully traverse this rejection. The claims do not require that a specific molecule or molecule complex be detected. Therefore, the Examiner is adding meaning into the claims that is not apparent from the claims or the specification. The addition of part (d) in claim 63 should resolve the alleged technical problem with the claim.

The Examiner's objections to claim 22 in paragraphs 10 and 11 are moot in view of the cancellation of claim 22.

In paragraphs 12 and 13, the Examiner again objects to claim 23 for not reciting active steps. In response, Applicants point out that the method steps are clearly set forth in claim 63 upon which claim 23 depends. Claim 23 simply qualifies one of those steps. In view of this explanation, Applicants respectfully request the Examiner to withdraw this rejection.

The Examiner asserts that claim 33 is confusing because it is not clear how to monitor "self-assembly" of a nucleic acid or antibody. This issue as been addressed above.

The Examiner rejects claim 37 for reciting "which." Applicants request clarification of this rejection.

The Examiner rejects claim 38 for reciting "contacts with a surface." This rejection is moot in view of the amendment to claim 37.

The Examiner asserts there is no antecedent basis for "second molecule" in claims 39, 40, 42 and 44. Applicants assert the amendment to claim 37 provides the antecedent basis.

The Examiner rejects claim 43 for a lack of antecedent basis for "second polynucleotide." Applicants assert that the antecedent basis for this term can be found in claim 42.

The Examiner states that in claim 44, there is no antecedent basis for "the first, second and third polynucleotides." Applicants assert that the antecedent basis for these terms can be found in claims 42 and in claim 44 itself.

The rejection to claim 51 is rendered moot with the cancellation of this claim.

In view of the above explanations and amendment, Applicants respectfully request the Examiner to withdraw the rejections under 35 USC § 112, second paragraph.

Applicants have amended claim 24 to add that the phase angle is "of the current" in response to the Examiner's suggestion during the interview.

With regard to claim 25, during the interview the Examiner expressed confusion about how the binding occurred and whether the detected molecule would be denatured. In response, Applicants point out that "diffusion" is discussed in the application at page 9, lines 4-14. Diffusion can be done by using chemically/physically related changes in concentration, and by applying an electric potential which produces a diffusion gradient. The binding may be physical (adsorption, Van-der-Waals forces) or chemical. Very common is that a protein/polypeptide or a nucleic acid having a thiol group (-SH) binds to a gold electrode via an Au-S-protein/polypeptide/nucleic acid link. However, this is of course only one example. In these cases, no further binding compound is necessary. The Examiner's concern about a denaturation is misplaced. If a protein is bound in such a way to the electrode, it will change the current or potential of the electrodes, and that is what is measured, independent of the further fate of the molecule.

III. Prior Art Rejections

The Examiner has rejected claims 21-25, 27-34, 37-40, 42-55 and 59-62 under 35 USC § 102 and 35 USC § 103 over U.S. Patent No. 5,653,939 ("the Hollis patent"). Applicants traverse this rejection as it may be applicable to the new or amended claims.

Applicants assert that Hollis does not teach all of the features of the claimed invention. For instance, it does not teach electrodes that are either a layer on a planar insulating support material, or that are incorporated in said planar insulating support material. Rather, Hollis teaches "microwells." This distinction is relevant because planarity of the present invention permits faster diffusion of molecules and greater

accessibility to targets, when complexes are studied. This planarity is nowhere taught or suggested in Hollis. Additionally, Hollis does not teach or suggest the spacing of the electrodes, as recited in the new and amended claims.

In view of this explanation and the above amendments and new claims, Applicants respectfully request withdrawal of the above prior art rejection.

CONCLUSION

In view of the above amendment and remarks, Applicants respectfully request that all objections and rejections be withdrawn and that a notice of allowance be forthcoming. The Examiner is invited to contact the undersigned attorney for Applicants at 202-919-2142 for any reason related to the advancement of this case.

Respectfully submitted,

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VERSION WITH MARKINGS TO SHOW CHANGES MADE

23. (Amended) A method according to claim [21] 63, wherein the **measuring of the** changes in current or potential **is performed** [are measured] using impedance spectroscopy.

24. (Amended) A method according to claim [21] 63, wherein the changes in current or potential are measured independently of time, as a function of time or as a function of the phase angle **of the current**.

25. (Amended) A method according to claim [21] 63, wherein the changes in current or potential are caused by diffusion or binding of the **first** molecule or **first** molecular complex to the ultra-microelectrode array.

27. (Amended) A method according to claim [21] 63, wherein the alternating electric field comprises, is superimposed, or excited with a direct-current component.

31. (Amended) A method according to claim [21] 63, wherein the **first** molecule or **first** molecular complex binds to **a binding compound on** a surface of the electrode structures.

32. (Amended) A method according to claim [31] 63, wherein the **first** molecule or **first** molecular complex binds to the surface of the electrode structures via physical or chemical binding.

33. (Amended) A method according to claim [31] 63, wherein the **first** molecule or **first** molecular complex binds to the surface of the electrode structures via self-assembling.

34. (Amended) A method according to claim 31, wherein the **first** molecule or **first** molecular complex binds to **the binding compound on** the surface of the electrode structures via electropolymerization.

37. (Amended) A method according to claim [21] **63**, comprising [wherein the electrode structures are layered with a substrate which is bound to an antigen or a nucleic acid molecule, said antigen or said nucleic acid molecule capable of binding to the molecule or molecular complex to be detected] **a second molecule, the second molecule being selected from the group consisting of antigens and nucleic acid molecules, that binds to the first molecule or first molecular complex to be detected, wherein the second molecule is bound directly to said ultra-microelectrode array or is bound via a binding compound, and whereby the binding between the second molecule and the first molecule or first molecular complex to be detected is capable of causing the changes in current or potential between the electrode structures**.

38. (Amended) A method according to claim 37, wherein the second molecule [molecular layer contacts] **binds a binding compound on** [with] a surface of the electrode structures.

40. (Amended) A method according to claim 37, wherein the second molecule comprises an antigen, and wherein the **first** molecule or **first** molecular complex to be detected comprises an antibody.

42. (Amended) A method according to claim 37, wherein the second molecule comprises a first polynucleotide, and the **first** molecule or [molecule] **first molecular** complex to be detected comprises a second polynucleotide capable of binding to the first polynucleotide.

44. (Amended) A method [according to claim 37, wherein the second molecule comprises a first and second polynucleotides, wherein the molecule or molecule complex to be detected comprises a third polynucleotide, and wherein the first, second and third polynucleotides are capable of forming a triple helix] **of detecting a molecule or molecular complex in a sample, comprising:**

(c) **contacting the sample comprising a first molecule or a first molecular complex with a single ultra-microelectrode array, said**

ultra-microelectrode array comprising at least two electrode structures,

(d) producing an electric field between the electrode structures; and

(c) measuring changes in current or potential between the electrode structures, whereby the changes in current or potential are caused by the first molecule or the first molecular complex, wherein said first molecule or molecular complex is a third polynucleotide that hybridizes to a second polynucleotide that is hybridized to a first polynucleotide, wherein said first polynucleotide is bound to a binding compound on said ultra-microelectrode array; and

wherein each of said electrode structures is insulated from each other and is either a layer on a planar insulating support material or is incorporated in said planar insulating support material; and

wherein the spacing between the electrode structures is about 1 µm or less; and

wherein the electrode structures are arranged so closely next to one another that they approach the size of large molecule complexes.

45. (Amended) A method according to claim [21] **63**, wherein the ultra-microelectrode array comprises first molecular layer and a second molecular layer, wherein the first molecular layer contacts the second molecular layer, wherein the second molecular layer comprises a second molecule, wherein the second molecule is capable of binding to the molecule or molecular complex to be detected, and whereby the binding between the second molecule and the molecule or [molecule] **molecular** complex to be detected is capable of causing the changes in current or potential between the electrode structures.

46. (Amended) A method according to claim **[21] 63**, wherein a surface of the electrode structures comprises a layer of conductive material.

50. (Amended) A method according to claim [49] 63, wherein the insulating material is selected from the group consisting of silicon compounds, glass, ceramic and organic polymers.

53. (Amended) A method according to claim [52] 63, wherein the insulating material is selected from the group consisting of silicon oxides, nitrides, ceramic and plastics.

54. (Amended) A method according to claim [21] 63, wherein the electrode structures are arranged [to have] as a multi-layer structure with each layer insulated from one another.

59. (Amended) A method according to claim [26] 71, wherein the changes in current or potential are measured sequentially, in parallel or simultaneously.

62. (Amended) A method according to claim [47] 63, wherein the noble metal is selected from the group consisting of gold, platinum and iridium.

Please add the following new claims:

63. (New) A method of detecting a first molecule or a first molecular complex in a sample, comprising:

(a) contacting the sample comprising a first molecule or a first molecular complex with a single ultra-microelectrode array, said ultra-microelectrode array comprising at least two electrode structures;

(b) producing an alternating electric field between the electrode structures;

(c) measuring changes in current or potential between the electrode structures, whereby the changes in current or potential are caused by the first molecule or the first molecular complex; and

(d) detecting the presence of said first molecule or first molecular complex by observing said change in current or potential;

wherein said first molecule or first molecular complex is selected from the group consisting of nucleic acids, peptides and proteins; and

wherein each of said electrode structures is insulated from each other and is either a layer on a planar insulating support material, or is incorporated in said planar insulating support material and wherein the spacing between the electrode structures is about 1 µm or less; and

wherein the electrode structures are arranged so closely next to one another that they approach the size of large molecule complexes.

64. (New) A method according to claim 63, wherein the first molecule or first molecular complex is positioned in the gap by chemical binding, adhesion, or condensation reactions.

65. (New) A method according to claim 37, wherein said second molecule comprises an antibody, and wherein the first molecule or first molecular complex to be detected comprises an antigen that binds to said antibody.

66. (New) The method of claim 63, wherein a surface of the electrode structures comprises a layer of conductive material, said material being selected from the group consisting of a noble metal, a carbon material and both a noble metal and carbon material.

67. (New) The method of 63, wherein said ultra-microelectrode array is a noble metal or a carbon material or comprises said noble metal or carbon material.

68. (New) The method of claim 66, wherein said noble metal is selected from the group consisting of gold, platinum and iridium.

69. (New) The method of claim 67, wherein said noble metal is selected from the group consisting of gold, platinum and iridium.

70. (New) A method according to claim 63, wherein each of the electrode structures is a layer sufficiently thin that the electrode layer is substantially planar.

71. (New) A method according to claim 63, wherein the electrode structures are stacked, and comprise crossover points that are insulated from one another.

72. (New) A method according to claim 63, wherein the first molecule or first molecular complex is positioned in the gap between the electrode structures.

73. (New) A method according to claim 72, wherein the first molecule or first molecular complex is positioned by chemical binding, adhesion, or condensation reactions.